PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: Ekapot Bhunachet, M.D.,PhD

Applicant No. 09/936,872

Title: "FLUORESCENCE ELECTRONIC ENDOSCOPIC SYSTEM"

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Reply to the final action filed on June 14, 2007, and interview summary on August 10, 2007

Michael Rozanski
Examiner
Art Unit 3768

Dear Mr Rozanski,

As suggested by examiners, an affidavit that substantially and accurately demonstrates that the results of my invention were not predictable has been submitted.

As mentioned in the affidavit, optical fiber-scopes have typically been used for fluorescence observation, though they provide poor images of inferior quality compared with those of electronic endoscopes (or video-endoscopes) (Reference 14, page 886). It is believed that a conventional miniaturized CCD can hardly detect fluorescence lights, which are extremely weak especially in the case of auto-fluorescence (References 12 and 13). This is because the smaller the CCD, the less sensitive it is to light (Reference 14, page 888). MacAulay et al., themselves, mentioned in US 5,827,190 (Palcic B, MacAulay C, et al.) (col. 2, lines 14-18) that, ***"Prior art endoscopes have been developed that permit the image sensor to be located at the tip of the endoscope probe, however, in general, this endoscope equipment is intended***

for collecting reflected light and is not suitable for reliably capturing faint fluorescence images". Therefore it can be seen that nobody, including MacAulay et al., would ever have anticipated that a commonly used electronic endoscopic system using a black and white CCD in combination with a rotary color filter wheel with red, green and blue filters to obtain a white light color image could be turned to an excellent fluorescence electronic endoscopic system, simply by placing a barrier filter in front of the black and white CCD to eliminate excitation light. It should also be noted that the technique of using black and white CCDs in video-endoscopes is older than the technique of using color CCDs in video-endoscopes (Reference 14, page 888) and thus was not seen as a progressive technology from which could be expected superior results.

MacAulay et al'660 mentions methods of superimposing the integrated auto-fluorescence image and the remittance light image. However, there is an understandable reason that their method does not work. The fluorescence endoscopic system, LIFE (Light-Induced Fluorescence Endoscopy) manufactured by Xillix Technologies (the Assignee of US 5,590,660 and US 5,827,190), Canada and Olympus Corp., Tokyo, Japan, can only provide a fluorescence image (References 4-7). In the LIFE system, the auto-fluorescence is divided into green and red channels, instead of being integrated (Reference 4, page 235).

It can reasonably be assumed that Xillix Technologies had tried to integrate auto-fluorescence to one channel as described in MacAulay et al'660, but failed. The reason for this failure was probably the use of color CCDs. For example, in the forth embodiment of MacAulay et al'660 (col. 9, lines 53 to col. 10, line 10), a color CCD is placed at the tip of an endoscope with a barrier filter to exclude the blue excitation light and permit passage of green and longer wavelengths. Since the image sensing means is a color CCD, the auto-fluorescence, composed mainly of green light but also red light (Fig. 1a-d, MacAulay et al'660), will be sensed by both green and red channels.

In my invention where a black and white CCD is used, the fluorescence passing through the barrier filter is, as total, sensed and converted to an

electric signal simply using the blue channel (unless pseudo-color technique is used). Using a black and white CCD with a barrier filter also provides another advantage over using a color CCD with a barrier filter: remittance green and red lights, which are the same colors as the fluorescence, can be used to pick up the background images with the green and red channels, and these can be superimposed with the fluorescence image sensed by the blue channel. As a result, the image obtained is composed of three colors.

By contrast, if a color CCD is used with a barrier filter, the blue remittance light cannot be used since the barrier filter eliminates all blue light. The green remittance light cannot be used either, because it will be sensed by the same green channel as the majority of auto-fluorescence. Also, it will be impossible to distinguish the fluorescence image from the background image. Therefore, using a color CCD with the barrier filter can provide images made up of only two colors.

MacAulay et al'660 mention the use of three different channels (col.8, lines 21-35) in an additional modification of the apparatus of their third embodiment. Though it has never proven successful, this apparatus can only function when using two cameras attached at the end of a fiberscope, together with a set of dichroic mirrors and filters. This technique should be considered as different from my invention, which functions with a black and white CCD placed at the tip of an electronic endoscope with a barrier filter.

As described in detail in the affidavit, examining the evolution of various systems in the field of fluorescence endoscopy, it is evident that my invention provides results, which had not, and indeed could not, have been anticipated by the endoscopic research community. The techniques used in combination with my invention, i.e. as described in claims 35-40, are therefore not obvious but rather represent simple but profound innovations in the field of endoscopy.

In view of the above, claims 35-40 in this application, together with claim 34, which is now allowed, are believed to be in immediate condition of allowance. Accordingly, the examiner is respectfully requested to pass this application to issue.

If, for any reason, the examiner finds the application other than in condition for allowance, the applicant requests that the examiner contact Mr. Paul Sadler, a friend who is acting as the interpreter and translator for the Applicant, at his contact number (011-81-29-851-8038) or by e-mail at paulsadler@tsukubagrace.org to discuss any steps necessary to place the application in condition for allowance.

If there are any fees required in relation to this communication, please inform the applicant at the fax/phone number 011-81-29-851-3721.

Respectfully submitted,

Date: September 13, 2007

By: Ekapot Bhunachet

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CERTIFICATION OF MAILING

I hereby certify that this correspondence, together with an affidavit, are being deposited with EMS mail in an envelope addressed to Examiner Michael Rozanski, Art Unit 3768, UNITED STATES PATENT AND TRADEMARK OFFICE, P.O. Box 1450, Alexandria, VA. 22313-1450, on September 13, 2007.

Name of applicant: Ekapot Bhunachet

Date of Sig.: September 13, 2007

Signature: Ekapot Bhunachet

Affidavit

My name is Hideki Toyoda, M.D., PhD., an associate professor in the Mie University School of Medicine, Japan. I use endoscopes both in routine work and research. My publications in the field of endoscopy include the following:

1. Toyoda H, Rubio C, Befrits R, Hamamoto N, Adachi Y, Jaramillo E. Detection of intestinal metaplasia in distal esophagus and esophagogastric junction by enhanced-magnification endoscopy. Gastrointest Endosc. 2004 Jan; 59(1):15-21.
2. Tanaka K, Toyoda H, Inoue H, Hamada Y, Aoki M, Kosaka R, Takamura M, Imoto I.. Depressed-type early duodenal carcinoma (carcinoma in situ) observed by enhanced magnification endoscopy. Endoscopy. 2007 Apr 18; [Epub ahead of print]
3. Tanaka K, Toyoda H, Kadowaki S, Hamada Y, Kosaka R, Yamanaka M, Imoto I. Use of proton pump inhibitors may cause squamous epithelial masking of intramucosal carcinoma in Barrett's esophagus. Endoscopy. 2007 Apr 18; [Epub ahead of print]
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Introduction

As an expert in the field of endoscopy, I would like to make an affidavit stating that the fluorescence electronic endoscopic system developed by Dr. Ekapot Bhunachet provided results that could not have been anticipated or predicted based upon research that had been done up until the point of this invention. Furthermore, these results constitute a major breakthrough in

endoscopy image quality, delivering far superior image clarity to any endoscopy systems that preceded it. A summary of the history of fluorescence endoscopy development demonstrates this.

Weaknesses of Previous Endoscopic Systems

LIFE system (Xillix Technologies, Canada) was the first auto-fluorescence (AF) endoscopic system to be used widely in clinical studies. Since 1993, there have been many studies reporting that LIFE bronchoscopy can improve detection rate of early lung cancers (Reference 1, Table 4), (References 2-3). LIFE-GI (Light-Induced Fluorescence Endoscopy for GastroIntestine, Xillix Technologies, Canada and Olympus Corp., Tokyo, Japan) was reported to be able to distinguish most gastrointestinal tumors (Reference 4). However, LIFE has some serious weak points. The system by using a fiber-optic endoscope and a heavy image intensifying camera unit (with two image intensifier CCD cameras inside) attached to the eyepiece failed to provided sufficient image quality and maneuverability, and was not feasible for general use in the era of high resolution video-endoscopy (or electronic endoscopy) (References 4-7). The specificity of LIFE in detecting early esophagogastric cancers (Reference 5), Barrett's esophageal cancers (Reference 6) and lung cancers (Reference 7) is found to be low. There are two possible explanations for these results. First, the algorithm used to construct the AF image accounts only for the ratio of red to green AF and does not incorporate information from the reflected light. Second, the LIFE system included fiberoptic endoscopes, which provide a poor white-light image compared with currently available, high-quality video-endoscopes (Reference 6, page 679).

In 1999, there were two newer AF systems introduced, the D-Light System (Storz, Tuttlingen, Germany) (References 1, 8 and 9) and the SAFE 1000 (Pentax, Asahi Optical Tokyo, Japan) (References 1, 10 and 11). These proved to be cheaper and more

convenient in their handling. However, these two newer AF systems, like the LIFE system before them, used fiber-optic endoscopes, and the images obtained remained dim, showing little appreciable improvement on the images produced by the LIFE system (Reference 9, Figures. 1 and 2), (Reference 11, Figures. 2 and 3).

Industry Fails to Resolve System Weaknesses

As a result of the poor clarity in images developed using this technology, for years there had been strong demand from clinicians to develop a new generation of AF system based on video-endoscope technology.

As described above, optical fiberscopes have typically been used for fluorescence observation. It is considered that a conventional miniaturized CCD can hardly detect fluorescence lights, which are extremely weak especially in case of autofluorescence (References 12 and 13). This is because the smaller the CCD, the less sensitive it is to light (Reference 14, page 888). Interestingly, in 1993, there had been a report of a study using a fluorescence video-endoscopic system developed by Olympus (Tokyo, Japan) (Reference 15). This system, however, failed to demonstrate real-time changes of fluorescence on the surface of colonic mucosa. Electronic endoscopic images were sequentially recorded as digital data every second for two minutes after intravenous administration of fluorescent material. Each image had to be processed by computer in order to accentuate the change with fluorescence.

Breakthrough Fluorescence Endoscopic System

In 2002, Bhunachet et al. reported the first study using a fluorescence electronic (or video) endoscopic system able to

demonstrate real time changes of fluorescence on the surface of gastric mucosa after intravenous injection of fluorescein sodium (Reference 16). In this study, it was reported that fluorescein electronic endoscopy was useful in determining the extent within the mucosa of gastric cancers when this was obscure by standard endoscopic observation, and for detecting extremely early stage cancer that was not evident by conventional endoscopic observation. This fluorescence electronic endoscopic system was also able to observe the difference of auto-fluorescence emitted from an early gastric cancer and the surrounding normal gastric mucosa (Reference 16, Fig. 8F). Beside these impressive results, one of the most striking points of this study is that Bhunachet et al. themselves developed this fluorescence electronic endoscopic system using a conventional electronic endoscopic system (Olympus, Tokyo, Japan) commonly used, and especially widespread in Japan.

This system has a light source, with a rotary red/green/blue band-pass filter. With this light source, the mucosa is sequentially illuminated with red, green, and blue light. The reflected red, green, and blue light is detected by a black and white CCD and is converted to an electronic signal that is passed to the video processor, which is synchronized with the rotary filter. The processor electronically overlaps the red, green, and blue signals to produce a high-quality white-light image. The only items needed to turn the commonly used electronic endoscopic instrument to a fluorescence electronic endoscopic system are a thin-filmed barrier filter and a glass adjuster filter, which cost less than $30. In this study, the barrier filter was attached to the glass covering the objective lens of the endoscope with glue, and the adjuster filter was placed in the existing filter holder within the light source. The latter concentrating the blue light and cut off some portions of green and red light can be moved with a single touch in or out of light path of the lamp. The barrier filter eliminates the blue excitation light, but passes the fluorescence and green and red lights. The sequentially detected

image from fluorescence, green reflectance and red reflectance are integrated by the image processor into one image. The image obtained, therefore, has 3 spectral components: (1) total fluorescence in response to blue light excitation, (2) green reflectance light, and (3) red reflectance light (Reference 16, pages 567-569).

The images provided by the former fiber-optic fluorescence endoscopic systems, the LIFE, D-Light, and SAFE 1000 systems, are still relatively dim, despite the use of an intensified CCD camera. In the AF mode, LIFE (References 4-6) and SAFE 1000 (Reference 11) provide images composed of the fluorescence image, and D-Light combines the fluorescence image with some reflectance of blue light (Reference 1), (Reference 9, Figure 2). By comparison, the hand-made fluorescence electronic endoscopic system by Bhunachet et al. can produce bright, high quality images even in an organ with a large internal volume, such as the stomach, where images tend to be dim. Real time changes in fluorescence can be observed on a bright background. This is because the image provided by this fluorescence electronic system is a coalescence of the fluorescent image, picked up and amplified electrically by the blue channel, and the background image, which is picked up by the green and red channels, with light adjusted by the adjuster filter: that is, the image is constructed with all 3 colors. Because the fluorescence image and background image are received by different channels, it is easy to recognize the source of the fluorescence emitted on the background (Reference 16, page 569)

Scientific Community Recognizes the Advance Made Possible by a Fluorescence Endoscopic System

In 2005, there were 4 papers published using a newly developed auto-fluorescence imaging (AFI) video-endoscope system (Olympus, Tokyo, Japan) for detection of colonic (Reference 4),

esophaogastric (Reference 5), Barrett's esophageal (Reference 6) and lung cancers (Reference 7). This AFI video-endoscope system utilizes the same methodology as the system developed earlier by Bhunachet et al (Reference 16). It was reported that, compared to the LIFE system, this new AFI video-endoscope system, using a different algorithm (Reference 6, page 680), was more superior in its image quality and maneuverability. Photographs of the same lesions taken by the LIFE fiber-optic system and the AFI video-endoscopic system demonstrated evident superiority of the latter in both white light and auto-fluorescence modes (Reference 4, figure 7), (Reference 5, figure 4), (Reference 7, figure 3). There was lack of light for a view from a great distance, making diagnosis difficult (Reference 4, page 237). In contrast, images obtained by AFI provided better brightness and lesion margins were clearly evident (Reference 4, page 237).

While the LIFE system is bulky (Reference 9, page 396) having a heavy image intensifying camera unit (with two image intensifier CCD cameras inside) (Reference 4, figure 1) attached to the eyepiece of a fiber-optic endoscope, the AFI system has the same appearance (Reference 4, figure 2) and the same maneuverability as that of a conventional video-endoscope (Reference 4, figure 2), (Reference 5, page 522).

The diagnostic performance of the AFI videoendoscope system was reported to have higher specificity for the detection of colonic neoplasms than LIFE fiberscopic imaging (Reference 4, page 238). It was also reported that AFI may improve the detection of high grade dysplasia or early cancers in patient with Barrett's esophagus, while it had been proven in a randomized cross-over study that no increase in the rate of detection was found using LIFE (Reference 6, page 679). The AFI correctly diagnosed all superficial esophageal cancers without chromo-endoscopy. The LIFE system had a sensitivity of 91% and a specificity of 81% for detection of esophageal squamous cell carcinomas in patients

with head and neck cancer (Reference 5, page 527). The specificity for detecting pre-invasive bronchial lesions by AFI (83.3%) was significantly higher than that of LIFE (36.6%) ($p = 0.0005$) (Reference 6, page 307).

Relationship Between AFI and Fluorescence Electronic Endoscopic System of Bhunachet et al

From 2006, two fluorescence videoendoscopic systems have become commercially available, AFI (Olympus, Tokyo, Japan) and SAFE 3000 (Pentax, Tokyo, Japan). It is considered that the AFI video-endoscopic system is ***the first system*** that incorporates a high-resolution video endoscope and a combined fluorescence and reflectance imaging modality (Reference 17). The SAFE 3000 uses a diode laser light as the excitation light. The objective lens functions to eliminate the wavelength of excitation light, and passes only the fluorescence from the object to a *color CCD* (Reference 18, page 22). SAFE 3000 provides only fluorescence images without coalescence with reflection light images (Reference 18, Figures 3 and 4).

It should be pointed out here that the AFI system (Olympus, Tokyo, Japan) (References 4-7) used the same methodology as that of the hand-made fluorescence electronic endoscopic system developed by Bhunachet et al. based on a commonly used electronic endoscopic system (Olympus, Tokyo, Japan) (Reference 16) (Compare Figures 13 of Reference 16 with Figure 3 of Reference 4 and Figure 1 of Reference 5). ***Therefore, Bhunchet et al.'s system is actually the first system that incorporates a video endoscope and a combined fluorescence and reflectance imaging modality.***

In both systems, blue spectrum light is delivered for excitation, together with light in the green and the red spectra. A barrier filter is placed in front of a black and white CCD to eliminate the blue excitation light, but pass the fluorescence and green and red

lights. The sequentially detected image from fluorescence, green reflectance and red reflectance are integrated by the image processor into one image. The image obtained, therefore, has 3 spectral components: (1) total fluorescence in response to blue light excitation, (2) green reflectance light, and (3) red reflectance light. The only difference between the AFI system and the fluorescence electronic endosocpic system developed by Bhunachet et al. is that, in the AFI system, the image processor pseudocolors fluorescent image to green, the green reflectance image to red, and the red reflectance image to blue (Reference 5, page 522), but not in the latter.

Fluorescence Video-endoscopic System's Extraordinary Results Could Not Have Been Anticipated

At present, it is widely accepted that a newly-developed fluorescence video-scopic system with the same methodology as that of Bhunachet's system, the AFI system, is more superior than the fiberoptic LIFE system developed more than ten years ago both from the points of image quality and maneuverability (References 4-7). The algorithm used in this new fluorescence videoendoscopic system is different from that of the LIFE system (Reference 6, page 680). The LIFE system used to cost about 80,000,000 Yen (about $700,000). Now one can buy an AFI system for approximately 6,000,000 Yen (about $50,000). Clearly if anyone had anticipated a technical innovation of this kind a manufacturer would have stepped in with a new competitive product sometime during this ten year period.

Bhunachet et al. developed their fluorescence electronic endoscopic system merely by placing a barrier filter in front of the Black and White CCD of a conventional electronic endoscopic system commonly used prior to the introduction of the LIFE system (Reference 15). This fact is both remarkable and unexpected.

No one could have predicted that merely placing a thin-filmed barrier filter in front of the Black and White CCD of a commonly used electronic endoscopic system would turn it into an ideal fluorescence electronic endoscopic system. There was significant market demand for improvements in image quality experienced in existing technology and yet years passed without any resolution.

If anyone in the field had had such an idea, it would not have taken more than ten years after the development of the fiber-optic LIFE system for the video-endoscopic AFI system to be developed. Furthermore, if anyone could have predicted the extraordinary results of the AFI system, two fiber-optic fluorescence endoscopic systems exhibiting the same inherent weaknesses as LIFE, i.e. the D-Light and SAFE 1000 systems, would never have been developed in 1999.

Conclusion

Having witnessed the evolution of various systems in the field of fluorescence endoscopy, and experienced firsthand the dramatic benefits of the methodology developed by Bhunachet et al, I am forced to conclude that this system, impressive in its simplicity, has given birth to a significant innovation that had not, and indeed could not, have been anticipated by the endoscopic research community.

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Respectfully submitted

Date: September 12, 2007

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